



PATENT COOPERATION TREATY
PCT
INTERNATIONAL PRELIMINARY EXAMINATION REPORT
(PCT Article 36 and Rule 70)

Applicant's or agent's file reference ES/11229.193	FOR FURTHER ACTION		See Notification of Transmittal of International Preliminary Examination Report (Form PCT/PEA418)
International application No. PCT/CA 03/01183	International filing date (day/month/year) 06.08.2003	Priority date (day/month/year) 06.08.2002	
International Patent Classification (IPC) or both national classification and IPC C07C57/12			
Applicant UNIVERSITE LAVAL et al			
<p>1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.</p> <p>2. This REPORT consists of a total of 9 sheets, including this cover sheet.</p> <p><input checked="" type="checkbox"/> This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 807 of the Administrative Instructions under the PCT).</p> <p>These annexes consist of a total of 4 sheets.</p>			
<p>3. This report contains indications relating to the following items:</p> <ul style="list-style-type: none"> I <input checked="" type="checkbox"/> Basis of the opinion II <input type="checkbox"/> Priority III <input type="checkbox"/> Non-establishment of opinion with regard to novelty, inventive step and industrial applicability IV <input checked="" type="checkbox"/> Lack of unity of invention V <input checked="" type="checkbox"/> Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement VI <input type="checkbox"/> Certain documents cited VII <input type="checkbox"/> Certain defects in the international application VIII <input type="checkbox"/> Certain observations on the international application 			
Date of submission of the demand 14.01.2004		Date of completion of this report 17.01.2005	
Name and mailing address of the international preliminary examining authority:  European Patent Office - P.B. 5818 Patentlaan 2 NL-2280 HV Rijswijk - Pays Bas Tel. +31 70 340 - 2040 Tx: 31 651 epo nl Fax: +31 70 340 - 3018		Authorized Officer Delanghe, P Telephone No. +31 70 340-4119 	

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I. Basis of the report

1. With regard to the elements of the international application (*Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17)*):

Description, Pages

1-9, 11-22 as originally filed
10 received on 09.11.2004 with letter of 09.11.2004

Claims, Numbers

1-12 received on 09.11.2004 with letter of 09.11.2004

Drawings, Sheets

1/6-6/6 as originally filed

2. With regard to the language, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

These elements were available or furnished to this Authority in the following language: , which is:

- ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).
☐ the language of publication of the international application (under Rule 48.3(b)).
☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any nucleotide and/or amino acid sequence disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

- ☐ contained in the international application in written form.
☐ filed together with the international application in computer readable form.
☐ furnished subsequently to this Authority in written form.
☐ furnished subsequently to this Authority in computer readable form.
☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.
☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

- ☐ the description, pages:
☐ the claims, Nos.:
☐ the drawings, sheets:

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)).

(Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)

6. Additional observations, if necessary:

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IV. Lack of unity of invention

1. In response to the invitation to restrict or pay additional fees, the applicant has:

- ☐ restricted the claims.
- ☐ paid additional fees.
- ☐ paid additional fees under protest.
- ☐ neither restricted nor paid additional fees.

2. ☒ This Authority found that the requirement of unity of invention is not complied with and chose, according to Rule 68.1, not to invite the applicant to restrict or pay additional fees.

3. This Authority considers that the requirement of unity of invention in accordance with Rules 13.1, 13.2 and 13.3 is

- ☐ complied with.
- ☐ not complied with for the following reasons:

4. Consequently, the following parts of the international application were the subject of international preliminary examination in establishing this report:

- ☒ all parts.
- ☐ the parts relating to claims Nos. .

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes: Claims	1-11
	No: Claims	12
Inventive step (IS)	Yes: Claims	11
	No: Claims	1-10,12
Industrial applicability (IA)	Yes: Claims	1-12
	No: Claims	

2. Citations and explanations

see separate sheet

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Re Item IV

Lack of unity of invention

This examining division has found multiple (groups of) inventions in this international application.

1. Claims 1-11 define a method for preparing conjugated linolenic acid and new conjugated linolenic acids.
2. Claim 12 defines the use a selection of linolenic acids, obtained by the method defined above, in nutritional, cosmetic and nutraceutical applications.

The problem to be solved by the presently claimed subject-matter is the preparation of novel and known conjugated linolenic acids and its use in nutritional, cosmetic and nutraceutical applications. The solution to this problem as proposed in its broadest form is the provision of conjugated linolenic acids and their preparation. The technical feature which is common to the claimed subject-matter as a whole, is the conjugated linolenic acid structure.

The closest state of the art for the purpose of judging the unity of the presently claimed subject-matter consists of the following document, which describes the conjugated linolenic acid structure is document D3.

D3: WO 01/44485 A1, see abstract

Consequently this technical feature (the conjugated linolenic acid structure) has already been associated with the solution of the same problem in the prior art. As a result this technical feature can not be considered as the special technical feature within the meaning of Rule 13.2 PCT, which differentiates the claimed invention as a whole from the prior art. Consequently the application lacks unity within the meaning of Rule 13.1 PCT.

At this point the the applicant's attention is drawn to the fact that the formulation of a "use of compound" claim in terms of its preparation, does not change its category, nor does it render the claim novel and inventive.

Therefore, the claimed subject-matter has been split up as above on the basis of the

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method of preparation and the use of conjugated linolenic acids.

Finally, the applicant is alerted that since the examining division has found multiple inventions and although this has no influence on the examination in the PCT phase anymore, it is highly possible that when the applicant decides to file the invention in the European phase, objection to multiple inventions will be made.

Re Item V

Reasoned statement with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Documents

Reference is made to the following documents:

- D1: KASS, J.P.; BURR, G.O.: "Pseudo-eleostearic Acid" JOURNAL OF THE AMERICAN CHEMICAL SOCIETY, vol. 61, 1939, pages 3292-3294
- D2: US-B-6 316 645 (2001-11-13)
- D3: WO 01/44485 A (2001-06-21)
- D4: US 4 164 505 (1979-08-14)
- D5: The journal of biological Chemistry, Vol. 242, no. 24, 5686-5692, 1967.
- D6: US 4 393 043

The documents D5 and D6 were not cited in the international search report. However, the applicant has had the opportunity to react on these documents in the written opinion.

2. Subject matter

Claims 1-9 and 11 define a method for the preparation of conjugated linolenic acids, by heating a precursor mixture of the conjugated linolenic acid (e.g. a fat or an oil or a non-conjugated linolenic acid) with a (sodium) base in a (polyol) solvent, followed by an isolation step, using urea complexation or liquid chromatography. An enrichment step can also be included. Preferred compounds are 9Z,11E,15Z-, 9Z,13E,15Z-, 6Z,10E,12Z- and 6Z,8E,12Z-octadecatrienoic acid and its use in nutritional, cosmetic and nutraceutical applications (claim 12). The last compound is also claimed per se in claim 10.

3. Novelty

The document D1 discloses (page 3293, column 1, paragraph 3 - column 2, paragraph 2) the preparation of pseudo-eleostearic acid, 10,12,14-

octadecatrienoic acid (fully conjugated) by heating linseed oil fatty acids with potassium hydroxide in ethylene glycol, followed by crystallisation. The subject-matter of the claims 1,10,11 and 12 differs from this D1 in that urea complexation or liquid chromatography is used. In addition, neither 6Z,8E,12Z-octadecatrienoic acid, nor the preparation or the use of 9Z,11E,15Z- or 9Z,13E,15Z- or 6Z,8E,12Z- or 6Z,10E,12Z-octadecatrienoic acids are disclosed. The subject-matter of claims 1,10,11 and 12 is new over document D1 (Article 33(2) PCT).

The document D2 discloses (column 2, lines 10-28 and 51-59, examples 7-9 and figure 2) the preparation of conjugated linolenic acids by heating linolenic acid with Schlosser's base (n-BuLi/K-OtBu). Silver-ion impregnated HPLC is performed on the corresponding methyl esters as a means of analyzing the acid mixture. No conversion of the methyl esters back to the corresponding acids is disclosed. The base-treatment of linseed oil is not disclosed in D2. Also, neither the use of conjugated octadecatrienoic acid in nutritional, cosmetic and nutraceutical applications nor 6Z,8E,12Z-linolenic acid is disclosed in D2. The subject-matter of claims 1,10,11 and 12 is therefore new over D2 (Article 33(2) PCT).

The document D3 discloses (page 4, lines 17 - page 6, lines 8, page 8, lines 25-29, page 22, line 22 to page 23, line 11, page 25, lines 3-7, page 26, line 4-8, page 35, lines 26-29, page 42, line 30 to page 43, line 22, figure 3a and 3b) the conjugation of gamma-linolenic acid, especially starting from borage oil, using an alkali base in propylene glycol at 130 °C to 150 °C. HPLC or silver ion chromatography is used for analysis on the corresponding methyl esters. No conversion of the methyl esters back to the corresponding acids is disclosed. In addition, neither 6Z,8E,12Z-octadecatrienoic acid, nor the preparation nor the use of 9Z,11E,15Z- or 9Z,13E,15Z- octadecatrienoic acids are disclosed in D3. Also, linseed oil is not disclosed as feedstock. The subject-matter of claims 1,10 and 11 is new over document D3 (Article 33(2) PCT).

However, 6-Cis,10-trans,12-cis octadecatrienoic acids is disclosed in D3 as well as its use in a nutritional supplement. Regarding the subject matter of claim 12, it is noted that the addition that a compound is prepared by a novel and inventive process, does not necessarily render its use novel and inventive. The subject-matter of claim 12 is not new over document D3 (Article 33(2) PCT).

The document D4 discloses (column 1, lines 61-66, column 2, lines 29-33, claims 1-3 and 11) the conjugation of various oils, containing unconjugated linolenic acid

(not specified) using an alkali metal hydroxide, e.g. sodium hydroxide at temperatures between 200 °C and 370 °C, to be used as soaps. The subject-matter of the claim 1 differs from D4 in that the reaction mixture is not subjected to either urea complexation or liquid chromatography. In addition, neither 6Z,8E,12Z-octadecatrienoic acid, nor the preparation nor the use of 9Z,11E,15Z- or 9Z,13E,15Z-octadecatrienoic acids are disclosed in D4. The subject-matter of claims 1,10,11 and 12 is new over document D4 (Article 33(2) PCT)

4. Inventive step

- 4.1 Regarding the subject-matter of the novel claims 1-9, the present application does not meet the criteria of Article 33(1) PCT, because the subject-matter of claims 1-9 does not involve an inventive step in the sense of Article 33(3) PCT.

The document D3 is regarded as being the closest prior art to the subject-matter of claims 1-9 and discloses (see also above) the conjugation reaction of oils, containing unconjugated linolenic acids and the isolation of the conjugated linolenic acids. The subject-matter of claims 1-9 differs from this known D3 in that the reaction mixture, containing the conjugated linolenic acid is subjected to urea complexation or liquid chromatography.

The problem to be solved by this part of the present invention may therefore be regarded as an improved preparation and isolation process to obtain conjugated linolenic acids.

The feature liquid chromatography is merely one of several straightforward isolation possibilities from which the skilled person would select, in accordance with circumstances, without the exercise of inventive skill, in order to solve the problem posed. Therefore, as far as claims 1-9 relate to liquid chromatography, the subject-matter of claims 1-9 does not involve an inventive step.

- 4.2 However, the document D3 is also regarded as being the closest prior art to the subject-matter of claim 11. The subject-matter of claim 11 differs from this known D3 in that linseed oil is used and that different conjugated linolenic acids are also prepared.

Neither D3, nor any other document of the prior art discloses any process which solve the problem in the same way as the present application, namely by using linseed oil for the preparation of 9Z,11E,15Z- and 9Z,13E,15Z-octadecatrienoic

acids. Thus, given the teaching of the prior art, the skilled person would not consider solving the problem in the same way as the present application, and he certainly would not expect the improvement associated with the present application. Therefore, the solution proposed in claim 11 of the present application can be considered as involving an inventive step (Article 33(3) PCT).

4.3 However, regarding the subject-matter of the novel claim 10, the present application does not meet the criteria of Article 33(1) PCT, because as far as the claim 10 is new, the subject-matter of claim 10 does not involve an inventive step in the sense of Article 33(3) PCT, since no surprising effect has been shown for the novel compound of claim 10.

4.4 The document D3 is also regarded as being the closest prior art to the subject-matter of claim 12, and discloses (page 4, lines 17 - page 6, lines 8, page 8, lines 25-29, page 22, line 22 to page 23, line 11, page 25, lines 3-7, page 26, line 4-8, page 35, lines 26-29, figure 3a and 3b) the conjugation reaction of oils, especially borage oil for octadecatrienoic acids, containing unconjugated octadecatrienoic acid by using a base in propylene glycol solvent at temperatures between 130 °C and 165 °C and the isolation of the octadecatrienoic acid. 6-Cis,10-trans,12-cis octadecatrienoic acids is prepared and used in a nutritional supplement. The subject-matter of claim 12 differs from this known D3 in that different conjugated linolenic acids are also claimed.

The problem to be solved by this part of the invention may therefore be regarded as the use of an alternative conjugated linolenic acid in nutritional, cosmetic and nutraceutical applications.

The 9-cis,11-trans,15-cis octadecatrienoic acid has been disclosed in document D5 (see summary). It would be obvious to the person skilled in the art, namely when the same result is to be achieved, to introduce this compound 9-cis,11-trans,15-cis octadecatrienoic acid to be used as a nutraceutical, nutritional according to document D3, especially since no special technical effect has been shown for the use of the compounds in claim 12.

In addition, document D6 describes the use of conjugated linoleic and non-conjugated linolenic acids in cosmetics (see examples). The use of conjugated linolenic acids can therefore be seen as obvious alternatives and in the absence of a surprising effect, the subject-matter of claim 12 can not be considered as

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involving an inventive step.

The applicant's attention is in this respect again drawn to the fact that the formulation of a "use of compound" claim in terms of its preparation, does not change its category, nor does it render the claim inventive.

(9Z,12Z,15Z-C18:3 acid), from a natural source such as linseed oil, into 9Z,11E,15Z and 9Z,13E,15Z C18:3 acids, producing a mixture comprising approximately 30% of the conjugated linolenic acids. In a further embodiment, enrichment up to and over 40% is readily performed with urea crystallization. Moreover, the product is obtained in over 90% purity by simple preparative liquid chromatography. The products obtained include free fatty acids, and derivatives thereof, including, but not limited to esters, amides, salts as well as fatty alcohols. The method of the present invention produces the above mentioned conjugated trienoic acid with a high selectivity, in a short time period and under relatively mild conditions.

The present invention further relates to a method for preparing conjugated linolenic acids comprising the steps of:

- (a) blending a or a mixture of vegetable oils and/or fats including various concentrations of alpha or gamma and or both linolenic acids with a base to produce a reaction mixture;
- (b) recovering said conjugated linolenic acids from the reaction mixture, and
- (c) subjecting the reaction mixture to urea complexation or liquid chromatography.

Further scope and applicability will become apparent from the detailed description given hereinafter. It should be understood however, that this detailed description, while indicating preferred embodiments of the invention, is given by way of illustration only, since various changes and modifications within the spirit and scope of the invention will become apparent to those skilled in the art.

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WHAT IS CLAIMED IS:

1. A method for preparing conjugated linolenic acids comprising the steps of:

- (a) blending a or a mixture of vegetable oils and/or fats including various concentrations of alpha or gamma and or both linolenic acids with a base to produce a reaction mixture;
- (b) recovering said conjugated linolenic acids from the reaction mixture, and
- (c) subjecting the reaction mixture to urea complexation or liquid chromatography.

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2. The method as defined in claim 1, wherein said oils and/or fats are selected from the group consisting of amebia, basil, candelnut, flax (linseed), linola, gold of pleasure, hemp, mustard, perilla, soybean, canola, walnut, chia, crambe, echium, hop, kiwi, pumpkin, black currant seed oil, purslane seed oil, borage oil, and evening primrose oil as well as any other oil, wax, ester or amide which comprises free and/or derivatized linolenic acid.

3. The method as defined in claim 2, wherein said base is selected from the group consisting of sodium hydroxide, sodium alkoxylate, sodium metal, potassium hydroxide, potassium alkoxylate, potassium metal and strong base resins.

4. The method as defined in claim 3, further comprising isolating from said reaction mixture geometrical isomers of partially and/or fully conjugated isomers of said conjugated linolenic acids.

5. The method as defined in claim 1, wherein said blending is performed in a polyol solvent.

6. The method as defined in claim 5, wherein said polyol is selected from the group consisting of propylene glycol, glycerol and ethylene glycol.

7. The method as defined in claim 6, wherein said blending is performed at temperatures ranging from about 20°C to about 280°C over a period of time ranging from about 30 seconds to about 18 hours.

8. The method as defined in claim 3, wherein said liquid chromatography is reverse phase liquid chromatography.

9. The method as defined in claims 1 to 8, wherein said conjugated linolenic acids are selected from the group consisting of 9Z,11E,15Z-octadecatrienoic acid, 9Z,13E,15Z-octadecatrienoic acid, 6Z,8E,12Z-octadecatrienoic acid, and 6Z,10E,12Z-octadecatrienoic acid.

10. A 6Z,8E,12Z-octadecatrienoic acid of formula 1:



obtained by the method of claim 1.

11. A method for preparing 9Z,11E,15Z-octadecatrienoic acid and 9Z,13E,15Z-octadecatrienoic acid comprising:

- (a) blending linseed oil with a base to produce a reaction mixture; and
- (b) recovering said conjugated linolenic acids from the reaction mixture.

12. A use of conjugated linolenic acids selected from the group consisting of 9Z,11E,15Z-octadecatrienoic acid, 9Z,13E,15Z-octadecatrienoic

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acid, 6Z,8E,12Z-octadecatrienoic acid, and 6Z,10E,12Z-octadecatrienoic acid in nutritional, cosmetic, and nutraceutical applications, characterized in that the linolenic acids are obtained by the method of claim 1.

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